U 013891-8

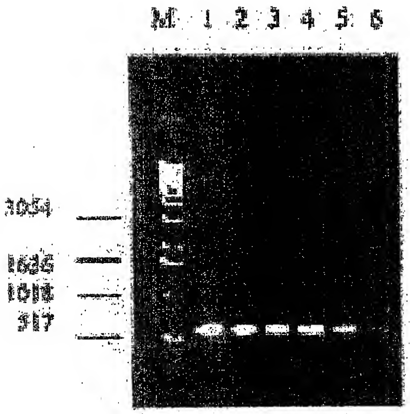

FIG. 1. Sensitivity of the PCR assay. Shown are the results of PCR amplification of the serially diluted *L. donovani* (DD8) DNA analyzed on agarose gels. DNA was extracted from parasite cultures and amplified as described in Materials and Methods. Lane M, 1 kb Ladder (Gibco BRL); lane 1, 10 ng of DNA; lane 2, 1 ng of DNA; lane 3, 10 pg of DNA; lane 4, 1 pg of DNA; lane 5, 10 fg of DNA; lane 6, 1 fg of DNA.

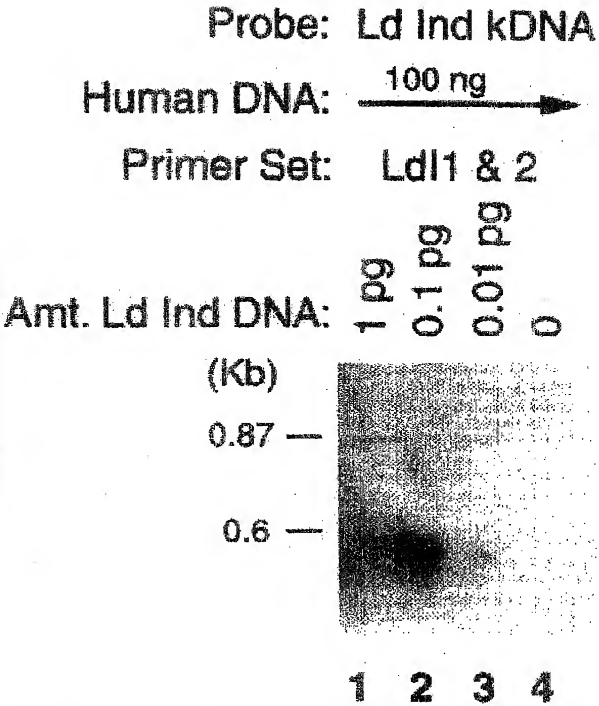

FIG. 2. Sensitivity of PCR amplification of *Leishmania* kDNA fol-lowed by Southern blot analysis. The PCR contained 100 ng of human genomic DNA and the indicated amount of total DNA from *L. dono-vani* DD8. The PCR product was probed with parasite kDNA and exposed for about 1 h. Lane 4 represents a PCR containing only hu-man DNA as a control.

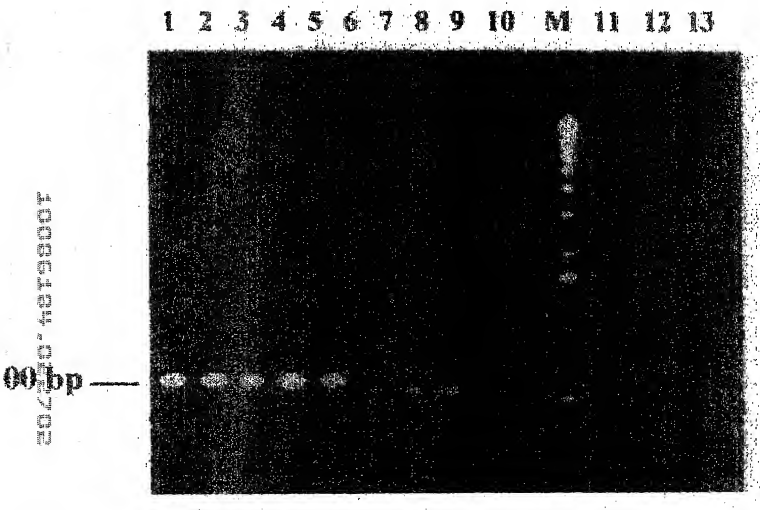

FIG. 3. Amplification of parasite DNA from various strains and olates of *Leishmania*. DNA (1 ng) isolated from parasite cultures was ubjected to PCR and analyzed. Lane 1, *L. donovani* AG83; lane 2, *donovani* DD8; lane 3, *L. donovani* HCB8; lane 4, *L. donovani* CB6; lane 5, *L. donovani* HCB 7 (PKDL origin); lane 6, *L. donovani* 8; lane 7, *L. donovani* WR684; lane 8 *L. donovani infantum*; lane 9, *tropica* WR683; lane 10, *L. major* LV 39, lane M, 1-kb ladder, lane , *Plasmodium*; lane 12, *M. leprae*: lane 13, *M. tuberculosis*.

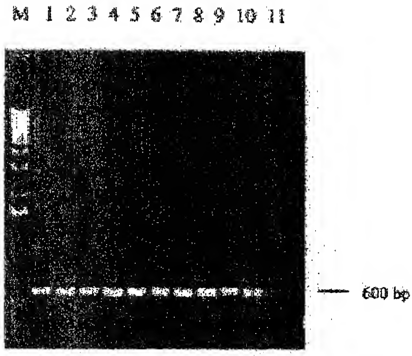

FIG. 4. DNA amplification from recent field isolates of KA and KDL. DNA (1 ng) extracted from cultures of parasite isolates was sed for PCR amplification. Lanes: M, 1-kb ladder; 1, KA-1; 2, KA-2; , KA-3; 4, KA-4; 5, KA-5; 6, PK-1; 7, PK-2; 8, PK-3; 9, PK-4; 10, PK-5; 1. isolate from a patient with cutaneous leishmaniasis.

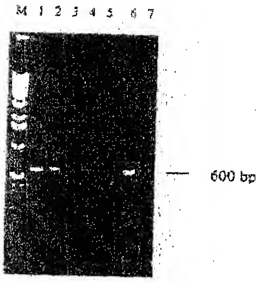

FIG. 5 PCR assay with clinical samples of KA and PKDL. DNA (100 ng) isolated from clinical samples was used for PCR amplification. Lane M, 1-kb ladder. lane 1, KA (bone marrow); lane 2, KA (blood); lane 3, malaria (blood); lane 4, tuberculosis (blood); lane 5, control from the area of endemicity (blood); lane 6, PKDL (skin lesion); lane 7, leprosy (lesion).

Fig 6. Sequence of PCR products with DNA isolated from *L. donovani* DD8 strain, isolates and clinical samples of KA and PKDL.

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